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FINAL REPORT

GENES FOR RUST RESISTANCE IN WHEAT

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PROJECT 97000031 - GENES FOR RUST RESISTANCE IN WHEAT

This project is somewhat unusual in that it is very broad and while some components have been completed others are ongoing. I have been awarded a new grant to continue the work. Thus, it is a bit unclear whether this is a Final Report or a Progress Report. Perhaps, it is a Final Progress Report. In any case, I will cover the components that have been completed and the progress that has been made on those that are continuing.

A. Abstract

The main objective of this project was to provide information about the inheritance of stem and leaf rust resistance in durum and bread wheat. This was done to a large extent by transferring individual genes from resistant parents into a susceptible variety by backcrossing. The lines carrying single genes were then tested with either 9 to 13 races of stem rust or 10 races of leaf rust to assess their resistance. In this way, a substantial number of previously unidentified genes for rust resistance have been isolated. Some were transferred from relatives of wheat. The lines produced can be used in wheat breeding, the identification of rust races and other wheat research.

B. Executive Summary

For almost 50 years, Canadian wheat breeders have prevented major rust damage to Canada's wheat crops. This has been possible because of research that has provided information about the inheritance of stem and leaf rust resistance and has identified useful genes for wheat breeding. The purpose of this research was to provide as much new information as possible about rust resistance and to develop lines carrying genes that will be useful to wheat breeders.

The main procedure used was to select varieties from many sources that appeared to have useful leaf or stem rust resistance and to transfer single genes for resistance from them to a common susceptible variety. For bread wheat, a line designated LMPG was developed specifically for this purpose. It is susceptible to all races of leaf and stem rust with which it has been tested. In some cases, resistance was transferred to durum or bread wheat from some of the wild relatives of wheat, particularly emmer wheat (wild durum).

Over a period of years, more than 500 lines carrying single genes for stem or leaf rust resistance have been developed. Most of them have been tested with sets of either stem rust or leaf rust races, whichever was appropriate. Initially, 9 races of stem rust

were used but this was increased first to 10 and then 13. Ten leaf rust races have been used. All of the data for stem rust were entered on a spreadsheet. The lines were sorted first according to the number of races to which they gave resistance and then by the infection types that they showed. The data were then analyzed to determine which lines carried previously unidentified genes for resistance and which would be most useful in wheat breeding. A number of lines gave resistance to all races of stem rust with which they were tested and should be useful in wheat breding.

A similar analysis of the data on leaf rust resistance will be done as soon as tests have been completed on the lines.

For a number of varieties, resistance proved to be quite complex. In theses cases additional genetic studies were done to determine the number of genes involved. Unexpectedly, in several cases it appeared that varieties carried genes that suppressed some of their genes for resistance. This was confirmed in a test involving seven, stem rust susceptible durum wheat varieties. Twenty crosses were made among the varieties and 14 segregated for resistance in the F2 generation. Six of the seven were crossed to a bread wheat and all six crosses segregated. These are quite surprising data.

Some of the results of these studies have been described in five papers, three that have been published and two in press. Seed of any of the material will be provided to anybody who requests it.

C. Technical Report

The primary objective of this research is to provide as much information as possible about the inheritance of stem and leaf rust resistance in wheat. The information can then be used by other wheat researchers, including wheat breeders. If wheat breeders are to continue to keep ahead of changes in the rust pathogens, they constantly need new genes for resistance with which to work. The current emphasis in wheat research is on biotechnology and molecular genetics. However, much of this work cannot be done without knowledge of the genetics of the character being studied.

1. Identification of New Genes for Rust Resistance in Wheat

a. Isolation and Identification of Genes

For many years prior to the initiation of this project, I had been transferring all of the identified genes for stem rust resistance in wheat to a common, susceptible genetic background, a line designated LMPG. LMPG was developed specifically for use in this work and it is suceptible to all races of leaf and stem rust with which it has been tested. The transfer of genes to LMPG involves backcrossing a line carrying a resistance gene to LMPG, usually five times. As far as I know, nobody else in the world is producing a set of lines of this type. I am currently working with scientists at the two North American

Rust Labs at Winnipeg and St. Paul, Minnesota, to have these lines used in the identification of stem rust races. This is one of many possible uses for them.

In addition, for many years I have been studying many sources of resistance and identifying possible new genes for resistance. These genes are then transferred to the same genetic background, LMPG. The backcross lines are tested with 9 - 13 rust races to determine the races to which each gene gives resistance. The rust data can then be used to decide if a new gene is in fact different than previously identified genes. Often several genes give resistance to the same races and appear to be the same but this can only be confirmed by crossing the lines.

When the project began, I had completed testing a set of 46 backcross lines and parents with nine races of stem rust, a second set of 158 lines with 10 races and a third set of 136 lines with 13 races. The more races that are used, the more likely it is that different genes can be distinguished. I have just completed the testing of the first set of lines with the four additional races. The second set has been tested with one of the additional three races and the last two will be done shortly. The development of a fourth set of 121 lines has been completed and so far they have been tested with 6 of the 13 races. The testing would be completed more quickly but for a lack of available growth chambers.

Similar, but less extensive work is being done for genes for leaf rust resistance. In this case, much of the work involves durum wheat. A first set of 186 lines and parents has been tested with 10 leaf rust races. A second set of 116 lines has been tested with nine races and testing will be completed shortly.

b. Analysis of Results

As a first step, all of the data on stem rust were entered on a spread sheet in a computer. The lines were then sorted, first according to the number of races they gave resistance to and then by the the infection types to each race on a 0 to 9 scale. This made it possible to compare all of the lines that gave resistance to only one race, or only two races, etc. The initial analysis has been completed using only the data for the nine races that all of the lines had been tested with. As soon as all of the lines have been tested with the 13 races, the complete analysis using all 13 races will be done.

As an example of the analysis, with 9 races there are 36 possible combinations of resistance to two races. Twenty-seven lines gave resistance to only two races and 9 of the 36 possible types actually occurred. The relatively low number of types (9) that occurred was partly because only some of the races were used in developing the backcross lines. The nine types of lines that gave resistance to different pairs of races clearly carried nine different genes for resistance. Lines that gave resistance to the same two races may or may not carry the same gene. For example, six lines gave resistance only to races 111 and C65. Based on their origins and infection types, it is probable that five different genes are

involved, but this can only be confirmed by intercrossing the lines. Two lines clearly carry the same gene.

The most valuable lines are those that have resistance to all races. Forty-nine lines including parents, some lines carrying previously identified genes and eight lines that appear to carry new genes were resistant to all races to which they were tested. A large amount of intercrossing of lines will be required to determine which lines carry different genes.

c. **Analysis of Individual Varieties**

Medea Durum Wheat - Medea durum has been used as a supplementary differential variety in identifying stem rust races at the Rust Lab at St. Paul, Minnesota. I set out to isolate its genes and transfer them to the bread wheat line, LMPG. The results proved to be complicated and eventually a genetic analysis was done as well. The results indicated that four genes for resistance were transferred from Medea to LMPG but that Medea probably carried four additional genes. Unexpectedly, three of the four genes that had been transferred to LMPG gave resistance to races to which Medea is susceptible. It was then shown that this was due to the presence in Medea of suppressor genes which suppressed some of its genes for resistance. The work has been published in the journal, Crop Science. Additional research is being done to transfer the othe four genes and to further study the action of the suppressors.

Triumph 64 - Triumph 64 is a winter wheat that has also been used as a supplementary differential at the Rust Lab in St. Paul. Its resistance to stem rust proved to be genetically very complex. The genes for resistance in Triumph 64 were transferred to LMPG. Twenty-two backcross lines were produced and tested with 10 races and additional genetic studies were done. The backcross lines appeared to carry at least nine genes but it is probable that Triumph carries some additional genes that were not isolated. Some of the lines were resistant to races to which Triumph 64 is susceptible. Again it was concluded that suppressors are involved. Four of the lines are resistant to all 10 races and should be useful in wheat breeding. Additional work is being done. The results have been published in a paper in Crop Science.

Thatcher - Thatcher has resistance to many of the old races of stem rust. Over the years, its resistance had proven to be genetically very complex and difficult to use in wheat breeding. I had been studying it for almost 40 years without much success. Finally, I crossed Thatcher with LMPG and selfed the progeny for four generations using a single seed from each plant in each generation. The resulting 169 lines were increased and tested with six rust races to which Thatcher is resistant. The results were very different for each race except for races 29-1 and 48A which gave very similar results. Genetics of resistance to a race was often very complex and in some cases suppressors appeared to be involved. Thatcher seems to carry at least 20 different genes most of which give resistance to only one of the six races. It is not surprising that Thatcher was difficult to use as a source of rust resistance in wheat breeding. To complicate matters further, even

though the Thatcher parent had been selected for uniform resistance to race 56, it proved to be genetically heterogeneous. The results have been published in the Canadian Journal of Plant Science. Further work is being done to isolate as many of the genes from Thatcher as possible and test them with the full set of 13 stem rust races.

Genes that control resistance in seedlings are usually effective in adult plants as well. To test this for Thatcher, the 169 lines from the cross, Thatcher X LMPG, were tested in separate field rust nurseries with races 56 and 11 of stem rust. The genes for seedling resistance proved to have some effect in the field. However, Thatcher clearly has additional genes that are effective only in adult plants. A brief paper on the results will appear shortly in the Canadian Journal of Plant Science.

d. Tests for Genes Suppressing Rust Resistance in Durum Wheat.

Since suppressors of stem rust resistance seemed to be unexpectedly frequent, a test was set up to determine their frequency in durum wheats. Seven durum wheat varieties susceptible to stem rust race 15B were intercrossed in all possible combinations except one (i.e., 20 crosses). The F_1 plants were all susceptible to race 15B. However, in the F_2 generation, 14 of the 20 crosses segregated for resistance. Usually, less than 1/4 of the plants were resistant. In four of the crosses, the seedlings showed a yellow, physiological flecking and this probably was involved in the resistance Six of the durum varieties were also crossed with the bread wheat, LMPG. The F_1 plants were all susceptible to race 15B but all six F_2 populations segregated. These results are very surprising and are being tested further in later generations. The results were presented in a paper at a Symposium on Durable Disease Resistance in The Netherlands in November, 2000.

A similar test has now been started in bread wheat using parent lines that are susceptible to both stem and leaf rust. Most of the F_1 seedlings have just been tested with race 15B of stem rust and all were susceptible. When these plants have been harvested, the F_2 progeny will be tested with both leaf and stem rust to see whether the crosses segregate for resistance to either type of rust.

e. Gene *Sr6* for Stem Rust Resistance

The gene *Sr6* is interesting because it is very temperature sensitive and its dominance changes depending on the cross and the race of rust used in the tests. I decided to try to determine whether the change in dominance in different crosses depended on the susceptible *sr6* alleles carried by different susceptible parents, different modifying genes they carried or their general genetic backgrounds. The test results indicated that the the susceptible parents used carried different *sr6* alleles that interacted with the dominant *Sr6* gene for resistance. Such an interaction between alleles is unusual but appears to be similar to a process called paramutation, first described in maize. In the crosses studied to draw this conclusion, anomalous segregations occurred in a number of families. I postulated that a transposable element is sometimes present at the *Sr6* locus

and results in the anomalous segregations. Additional tests are being done. A paper on the work is in press in the journal, Theoretical and Applied Genetics.

2. Genes for Leaf Rust Resistance in Durum Wheat

Susceptibility to leaf rust has generally not been a problem in durum wheat. However, in 1986 when leaf rust was heavy on the prairies, several varieties appeared to be more susceptible than in previous years and this caused some concern. Relatively little is known about the inheritance of leaf rust resistance in durum wheat. A program was begin to determine the inheritance of resistance in some prairie varieties and then in a group of resistant international varieties. Lines carrying single genes for resistance were produced by backcrossing to a susceptible durum, RL6089. One set of 186 lines and parents have been tested with a set of 10 leaf rust races. A second set of 116 lines has been tested with nine races and the test with the final race will be done shortly. Unlike the results with stem rust, many of the leaf rust resistant lines in the first set gave resistance to all 10 races. Consequently it will be much more difficult to analyze the data since so many lines appear to give the same results.

3. The Transfer of Leaf and Stem Rust Resistance to Wheat from Its Relatives

Many of the wild relatives of wheat have good resistance to stem and leaf rust. The resistance can be transferred to durum and bread wheat with varying degrees of difficulty, depending on how closely related the species are to durum and bread wheat. A graduate student of mine started a program to transfer resistance from a number of species, particularly emmer wheat, the wild form of durum. Stem and leaf rust resistance from the emmer wheats have been transferred into both RL6079 durum wheat and LMPG bread wheat by backcrossing. Some of the resulting lines have already been tested with sets of stem or leaf rust races and the remainder are in the tests that are now underway. Some of the lines look promising for use in wheat breeding.

Many durum wheat varieties have excellent resistance to leaf rust. Wheat breeders have often found it difficult or impossible to transfer this resistance to bread wheat, probably because the bread wheats carry genes that suppress the resistance. However, it is likely that different bread wheats will not carry the same suppressors. To test this, five leaf rust resistant durum wheats were each crossed with four susceptible bread wheats of diverse origin and then backcrossed to the bread wheats. For all five of the durums, resistance has been successfully recovered in at least one and usually more of the four backcrosses. Some of the derived lines are being tested with a set of leaf rust races to see if they will be useful in wheat breeding.

An attempt to transfer an excellent type of leaf rust resistance from diploid, einkorn wheat to bread wheat was unsuccessful. No backcross seeds were obtained on the F_1 hybrid. However, Dr. P Hucl was successful in obtaining seed from an einkorn cross made for another purpose. I am now transferring leaf rust resistance from his hybrid to LMPG by backcrossing.